IMAGE ANALYSIS FOR AUTOMATIC CLASSIFICATION OF MITOTIC CERVICAL CELLS

Raymond J. Madachy, M.S. *, *Yao S. Fu, M.D.* +

Medical Imaging Division
Department of Radiological Sciences, UCLA

+ Professor of Pathology
Department of Pathology, UCLA

ABSTRACT

To diagnose cervical cancer from biopsies, digital image analysis techniques and statistical discriminant analysis are used to identify mitotic cells in cervical biopsies since the frequency of mitotic cells is a marker of early cancer. No such research applied to cervical biopsies has been described in the literature to date.

Parameters extracted from each cell include areal and geometrical measures, optical density statistics, bending energy, and various statistical texture features.

The discriminant analysis attempts to classify mitotic cells from those in interphase on the basis of the image analysis output data vectors using pre-scored classifications.

The first comprehensive trial of the imaging technique for a dataset of 32 cervical cells showed that 88% of the cells could be properly identified automatically.

A fast, accurate and robust expert imaging system is ultimately desired for the classification of cervical cancers from biopsies.

INTRODUCTION

It is desired to diagnose cervical cancer as early as possible for optimal treatment planning. One way to detect abnormal cell growth associated with cancer is to count the frequency of dividing cells in a section.

Even for an experienced pathologist, identifying mitotic cells is time-consuming, tedious and prone to error. Thereby an automatic classification method of identifying mitotic cells from those cells in interphase would be a powerful diagnostic technique.

Digital image processing and statistical analysis can be used for this classification task A feature of this approach is that by using the statistical analysis, the minimal amount of extracted data needed to obtain high accuracy will be found thereby reducing clinical computation time.

This research uses pre-scored specimens for the statistical discriminant analysis to discriminate between mitotic and interphase cells and thereby produce parameter classification bands for future specimens.

The feasibility and performance of various image analysis methods are being tested for transfer to a smaller, more user-friendly image workstation for clinical use by pathologists. In other medical diagnosis applications, similar expert systems have even outperformed their human counterpart and it is hoped that this technique could possibly increase the accuracy of diagnosis.

PREVIOUS EFFORTS

Eccles and Klevecz [3] analyzed temporal information from time-lapse video to identify mitotic Chinese hamster cells. Kaman, et al [6] counted mitoses in breast cancer sections and showed 63% of the mitotic cells properly identified and 5% of the non-mitotic cells remaining misclassified. No research has been reported for cervical biopsies in the area of mitotic cell recognition to date.

PATHOLOGY

Dysplasia is abnormal cell growth and is a precursor of invasive cancer. It is the goal of pathology to diagnose dysplasia as soon as possible so that relatively easy techniques of removing the abnormal growth can be administered.

The frequency of mitotic cells in a tissue section is an indicator of the degree of dysplasia. For the squamous cell types encountered for the cervical biopsies, the cell cycle lasts about 14 days while the mitotic phase is only 6-12 hours long. Therefore, at any snapshot time there is a very small chance of observing mitosis for any given cell.

From the above, it is seen that the number of cells in mitosis is a good proliferation marker for dysplasia.

During the phases of mitosis [7], chromosones become visible, they arrange at the equator, separate and eventually uncoil. The different appearances are the physical basis for the descriptive parameters described in the cell featuring section.

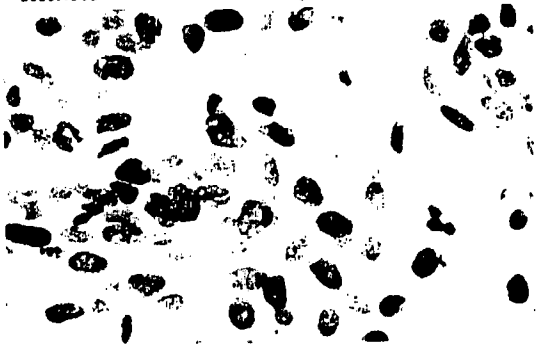

Figure 1: Cervical Biopsy

0372--IEEE ENGINEERING IN MEDICINE & BIOLOGY SOCIETY 10TH ANNUAL INTERNATIONAL CONFERENCE
CH2566-8/88/0000--0372 $1.00

METHOD

Cervical tissue sections of 5 microns thickness were taken as a routine histological procedure for this analysis. Additionally, single cell suspensions were prepared for simplifying the initial processing tests so that the cells in the image are not touching or overlapping. Photographs were then taken of the specimens under a Leitz microscope. Figure 1 shows a cervical biopsy.

These photographs are then digitized to 512*512 8-bit picture elements (pixels) with an Eyecom digitizing camera connected to a Gould/Deanza IP8500 image processor on a VAX host.

The analysis program uses a pre-specified density threshold and then searches each image for continuous contours based on the threshold. Small artefacts are eliminated by discriminating against those objects smaller than a fixed size.

To separate touching cells, this effort has used shrinking and expanding. Specialized morphological processing software optimized for the Gould image processor [9] is being modified for this purpose. These operations are optimized and operate at video rates in the digital video processor of the image processor. This processing is not necessary for the single-cell suspensions.

CELL FEATURE ANALYSIS

In this study various descriptors of cell images are computed including area, shape, density and statistical texture features [2] [4] [8].

From a cell contour which consists of coordinate pairs, the perimeter and area are obtained. The mean and standard deviation (SD) refer to the pixel density statistics enclosed within a cell contour.

SHAPE

The following morphological parameters are used to describe cell shape in this study (where P = perimeter, and A = area):

perimeter to area ratio = P / A

form factor (FF)= $P^2 / (4*\Pi*A)$

bending energy (BE) = $(1/P) * \sum_{i=1}^{N} R^2(i)$

where R(i) is the curvature at each point i in the outer cell contour given by

$$R(i) = \Delta\Theta / \Delta p$$

where $\Delta\Theta$ is the angle between segments of the cell contour and Δp is the average length between the segments. The angle Θ is defined as the change in direction along the contour in the local pixel neighborhood, P is the perimeter and N is the number of coordinate points along the complete contour [2].

When normalized with respect to perimeter or area, bending energy has been shown to be an effective and significant shape factor for classifying cell images and was found to be an important parameter in the statistical analysis performed here. This is due to the jagged edges exhibited by the mitotic cells.

TEXTURE

In this implementation, the region within the cell boundary is partitioned into three subregions based on density statistics whereby the black and white pixels are defined as those whose optical density is 15% greater or less, respectively, than the mean value for the whole cell. The black and white regions are eroded by two pixels along their periphery and then dilated so that only regions larger than 2 μm in diameter survive the operation. Figure 2 shows the texture regions in a cell.

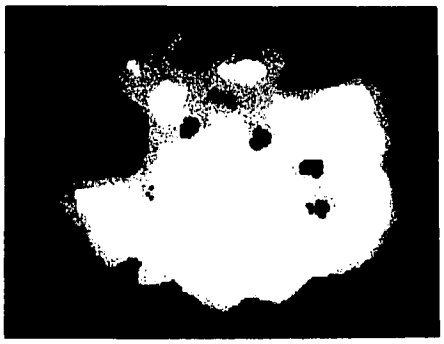

Figure 2: Cell With Texture Regions

From the resulting regions, the following parameters are calculated:

condensation = $A_{RB}+A_{RW} / A_T$
granularity = A_B+A_W / A_{RW}
clearing = A_{RB} / A_T
clumping = A_{RW} / A_T

where
A_T = total area enclosed in cell,
A_B = black pixel area before erosion and dilation,
A_W = white pixel area before erosion and dilation,
A_{RB} = remaining black area after erosion and dilation,
A_{RW} = remaining white area after erosion and dilation.

CELL CLASSIFICATION

An evaluation of the variance, correlation and class-separating power of the different parameters is accomplished with a stepwise discriminant analyis as implemented in the BMDP library developed at UCLA.

The stepwise discriminant analysis used in this effort chooses those parameters found to be most useful in setting the mitotic cells apart from other cells. The variables used to compute the linear classification functions are chosen in a stepwise manner.

RESULTS

In the first trial of the cell featuring, 18 mitotic cells and 14 non-mitotic cells were digitized for analyis. Statistical values such as F factors, P values and chi-square values were obtained for all the calculated parameters.

The results from the discriminant analysis were very encouraging. Using only the gray level standard deviation and bending energy, 81.2% of the cells could be classified. When the additional

parameters of log(area), perimeter, form factor and clumping are used, 87.5% of the cells can be identified. This is already better performance than that obtained in reference [5]. Tables 1 and 2 show a brief summary of the statistical classification results.

TABLE 1: PARAMETER STATISTICS

	INTERFACE		MITOSIS	
parameter	χ	σ	χ	σ
SD	26.83	10.32	38.75	6.52
CLEAR	0.41	0.08	0.50	0.08
MEAN	92.88	29.83	114.66	26.22
BE	2.33	0.78	2.79	0.92
PERIM	317.10	52.83	343.12	89.87
FF	1.39	0.52	1.47	0.33
CLUMP	0.34	0.08	0.33	0.06
AREA	3.77	0.14	3.79	0.21
PERIAREA	0.06	0.02	0.06	0.02
HET	0.75	0.14	0.83	0.07

TABLE 2: FINAL CLASSIFICATION SUMMARY

paramcters used: SD, BE, CLUMP, FF, PERIM, AREA

	interface	mitosis	% correct
interface	12	2	85.7
mitosis	2	16	88.9
total	14	18	87.5

WORK IN PROGRESS

In order to verify the accuracy of the technique and get better statistics, approximately 200 more cell images from specimens are being readied for digitization and analysis. Additionally, the tissue sections will be used instead of single cell suspensions. This will require shrinking and expanding of contours to separate the touching cells.

Additional classification features will be calculated and added to the database to gauge their usefulness in the task. Eventually, there will be an optimization made relative to the number of calculated parameters versus execution speed and classification confidence.

The main elements envisioned for this project in the final configuration are a conventional light microscope and a microcomputer with an image digitizing board. Eventually, such an automation garnered from these techniques could be used as part of an interactive expert pathological system employing artificial intelligence and neuron network parallel computing architecture.

SUMMARY

Algorithms have been developed for identifying mitotic cells in cervical biopsy sections. This unique capability can be used for automated pre-screening of patients and therefore reduce the physician's workload.

In the first trial of this technique, 88% of the mitotic cells were properly identified. Using a greater number of calculated parameters and/or refining current algorithms in further trials, it is likely that 95% or more of the cells can be identified. Additionally, the minimum amount of computations required to gain such accuracy will be found from the statistical analysis.

The cell analysis techniques will eventually be coded into a usable system for pathologists analyzing cervical biopsies. The powerful algorithms developed here can be particularly useful for general diagnosis and research of cancer in other tissue types as well.

REFERENCES

1 Bowie JE, Young IT: *An analysis technique for biological shape II.*. Acta Cytology 21:739-746, 1977.

2 Brugal G: *Image analysis of microscopic preparations.* Methods and Achievements in Experimental Pathology . vol. 11, pp. 1-33, 1984.

3 Eccles BA, Klevecz RR: *Automatic digital analysis for identification of mitotic cells in synchronous mammalian cell cultures.* Analytical and Quantititave Cytology and Histology 2:138-147, 1986.

4 Faugeras OD: *Fundamentals in Computer Vision.* Cambridge University Press, Cambridge. 1983

5 Jennrick R, Sampson P: *Stepwise discriminant analysis.* BMDP User's manual. pp.519-537, 1981.

6 Kaman EJ, Smeujlders AWM, et al: *Image processing for mitoses in sections of breast cancer: A feasibility study.* Cytometry 5:244-249, 1984.

7 Lavia MF, Hill RB: *Principles of Pathobiology.* Oxford University Press, New York. 1975

8 Levine MD: *Vision in Man and Machine.* McGraw Hill, New York. 1985.

9 Trambert MA: *Cellular logic implementation on a generalized image processor applied to biomedical image processing.* Analytical and Quantititave Cytology and Histology 2:131-137, 1986.